

## Protocol for metabolic activity determination

Cell viability, as a result of various enzymatic activities in live cultured cells, can be measured by using reagents / substrates that convert into measurable products (particularly optical density, for e.g. WST-1, Roche diagnostics).

- As per manufacturer's recommendation, add WST-1 reagent directly to the wells already seeded with hydrogels by adding 20µl reagent to 200µl medium in the well (dilution 1:10).
- Perform a blank with hydrogels without cells.
- Incubate at 37°C; 5% CO<sub>2</sub>.
- Perform optical density measurements at 440 nm by spectrophotometry after 30 minutes, 1h, 2h, 3h and 4h of incubation. This is based on the enzymatic activity of the cells tested.
- The sample should be returned to the incubator (37°C; 5% CO<sub>2</sub>) between each reading.

It is possible to use other commercially available kits for determining the metabolic activity such as:

- MTT.
- MTS.